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ORIGINAL PAPER

Electrochemical determination of sertraline in pharmaceutical formulation and serum using a gold electrode in a pH 8.4 bicarbonate solution

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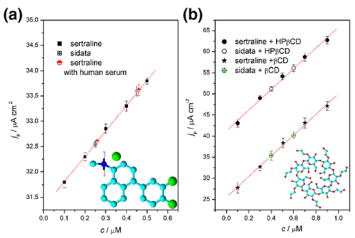
Abstract

The electrochemical characterization of sertraline at gold electrode was examined by cyclic voltammetry measurements (CV) in pH 8.4 bicarbonate buffer. Then Au electrode was evaluated for the quantitative determination of sertraline using square wave voltammetry (SWV). To enhance the sensitivity during the drug determination, (2-hydroxypropyl)-β-cyclodextrin (HPβCD) and β-cyclodextrin (βCD) inclusion complexes were employed. Using the proposed SWV technique, the anodic current peak was linear within a concentration range of 0.1–0.5 μM with a limit of detection (LOD) of 2.0×10^{-8} M and a limit of quantification (LOQ) of 6.7×10^{-8} M. In the case of inclusion complex of the sertraline with HPβCD, a good linearity range of 0.1–0.9 μM was obtained with a LOD of 2.6×10^{-8} M and a LOQ of 8.8×10^{-8} M. The gold electrode revealed the same linearity range for inclusion complex of the sertraline with βCD with a LOD and a LOQ being 2.6×10^{-8} and 8.6×10^{-8} M, respectively. Comparing the regression equations, it can be concluded that the sensitivity in the presence of inclusion complex can be up to 5 times higher. The applicability of the developed method was confirmed by the analysis of this drug in pharmaceutical formulation and in human serum spiked with sertraline standard. The comparison to HPLC method was successfully performed.

Graphic abstract



Keywords Cyclodextrines · Electrochemistry · Human serum · Pharmaceutical formulation · Voltammetry

Introduction

Sertraline hydrochloride in pharmaceutical formulation, sold under the brand name (Sidata, Zoloft, Lustral), is an antidepressant in the class of selective serotonin-reuptake inhibitors (Fig. 1) [1–3]. Serotonin is a neurotransmitter and is considered a happiness hormone. Numerous serotonin-reuptake inhibitors are effective and used in the treatment of depression, including sertraline. Sertraline is primarily used to treat clinical depression in adult patients, as well as obsession, panic disorder, and social phobia in adults and children. It is used orally and therapeutic doses of sertraline are 5–200 mg day^{-1} for four weeks, thus providing 80–90% inhibition of the serotonin transporter in the striatum [4]. Studies have shown an increase in the concentration

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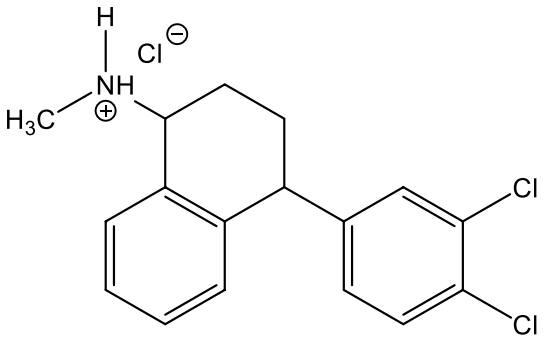

Fig. 1 The structure of sertraline hydrochloride

of amphetamines in the brain of rats pretreated with 5 mg kg^{-1} sertraline [5]. The drug is metabolized by the liver (N-demethylation) with a half-life of 26 h. It is excreted by the kidneys.

There is an increasing demand for rapid tests for the determination of trace concentrations of sertraline. In this context, different electrochemical methods showed a particular potential for practical applications in biomedical analyses. Square wave voltammetry (SWV) with electrodeposition at a hanging mercury drop electrode proved suitable for the determination of this drug in pharmaceutical formulations [6]. Using a pH 8.2 borate buffer for the samples containing 12% (v/v) methanol, a linear relationship between 2.33×10^{-7} and 3.15×10^{-6} M was obtained. A method using adsorptive voltammetry and a hanging mercury drop electrode flow cell can also been applied for the determination of sertraline in the concentration interval between 0.20×10^{-6} and 1.20×10^{-6} M [7]. Dermiş and Cay investigated the electrooxidation of this drug on a glassy carbon electrode (GCE) in pH 8 Britton-Robinson buffer-methanol supporting electrolyte using SWV [8]. A linear peak current–concentration relationship was found in the range from 0.04 to 0.8 mM. A rutin-modified GCE demonstrated the catalytic capability for the oxidation of sertraline, whereby the anodic current linearly related to the drug concentration in the range from 3.0 to 90.0 μM [9]. Shoja et al. described an innovative strategy for the determination of sertraline where nano-structured Ni(II)-levodopa film was electropolymerized on a GCE which was previously modified by gold nanoparticle-enriched multi-walled carbon nanotubes in an alkaline solution [10]. Namely, this nanostructure film was effective for the electrocatalytic oxidation of the drug and differential pulse voltammetry (DPV) was used for its determination in the range from 0.05 to 5.5 mM. On the basis of the electrooxidation of sertraline hydrochloride at a carbon paste electrode in presence of micellar medium, composed of a pH 7 Britton-Robinson buffer and Triton X-100, an analytical method was developed using square wave voltammetry for the determination of the drug [11]. Here, a linear relationship response was obtained from 1.99×10^{-7} to 1.38×10^{-5} M. A method for multiclass drug analysis using a biphenyl stationary phase and pulsed amperometric detection at a gold electrode was described by Muratt et al. [12]. This method enabled the determination of fourteen drugs as adulterants in dietary supplement samples, among which sertraline in the range from 3.00 to 70.00 mg dm^{-3}. Recently, Babaei et al. developed a GCE modified with a composition of Fe_3O_4@MCM-48-SO_3H and multi-wall carbon nanotubes for simultaneous determination of serotonin and sertraline in the presence of uric acid [13]. The DPV results showed a linear relationship between the concentration of sertraline and the anodic peak current in the range from 0.1 to 85 μM. A La_2O_3/Co_3O_4 nanocomposite modified screen-printed electrode enabled the determination of sertraline in a pH 7 phosphate buffer also using DPV [14]. In this case, the anodic peak current was increased linearly with the drug concentration in the range from 5.0 to 400.0 μM.

The importance of pharmaceutical application of cyclodextrins as drug carriers has been recognized for a long time [15]. Cyclodextrins, acting as hosts, are able to increase the solubility of the sparingly soluble compounds and thus, in the case of drugs, to enhance their bioavailability [16, 17]. Also, some other properties can be improved, such as resistance to light [18], organoleptic properties [19], as well as enhanced drug permeation [20]. Among diverse cyclodextrins, βCD has shown a significant ability in drug-sensing applications [21–23].

The aim of this work is to develop an electrochemical method using a gold electrode for quantitative determination of sertraline in pharmaceutical formulations and spiked serum. The method is based on the electrochemical oxidation of sertraline as the standard and its inclusion complexes (HPβCD, βCD) in a pH 8.4 bicarbonate buffer using SWV. The biologically relevant range of sertraline concentrations was examined and detailed statistical parameters of the obtained results are summarized and compared to the HPLC method.

Results and discussion

The use of CD complexes with pharmaceutically active compounds is attractive for the preparation of electrochemical sensors especially in pharmaceutical and biomedical analysis. It has been shown that cyclodextrin represents a non-electroactive host while pharmaceutical compounds are an electroactive guests [24–27].

In this work, the Au electrode was tested for the oxidation and determination of sertraline in a $NaHCO_3$ solution. The sertraline:CD inclusion complexes were prepared and used for sertraline electrooxidation ability improvement. It was proven that the complexation process of sertraline:CD

system is spontaneous and exothermic [28–30]. The most investigated systems with sertraline are complexes formed with βCD [31]. The stoichiometry of βCD or HPβCD complexes with sertraline is close to 1:1 with a negligible amount of 2:1 [28, 29] indicating that these CDs allow entrapment of one molecule of sertraline into the cavity. The high affinity of βCD in water is confirmed by the values of binding constants that vary from 5300 to 5900 M^{-1} depending on the used technique while the association constant for complex sertralin:HPβCD is 6530 M^{-1} [29]. Also, characterization of sertraline:βCD complex in solid state indicates a formation of 1:1 IC. It is also shown that possible interactions of the sertraline molecule with CD molecules include *N*-methyl unit as well as aromatic rings, especially fused aromatic ring [31, 32].

Figure 2 presents CVs for the oxidation of sertraline and its inclusion complexes with HPβCD as well as the voltammogram of the Au electrode in the blank solution for the sake of comparison. The electrochemical behavior of Au was described by taking into account adsorption processes, such as chemisorption of OH^-, which occurred in the potential region of -0.1 to + 0.3 V vs. SCE followed by the oxide formation at more positive potentials [33]. The onset potential for the oxidation of sertraline and its inclusion complex with HPβCD were correlated with formation of AuOH species. Also in the region of the AuOH layer, formation higher reaction currents were obtained with inclusion complexes of sertraline indicating the improvement of sertraline electrooxidation ability in the presence of HPβCD. The similar improvement of the oxidation abilities we reported for inclusion complexes of nifedipine and amlodipine also at Au electrode [34] as well as for arylazo pyridone dyes [35].

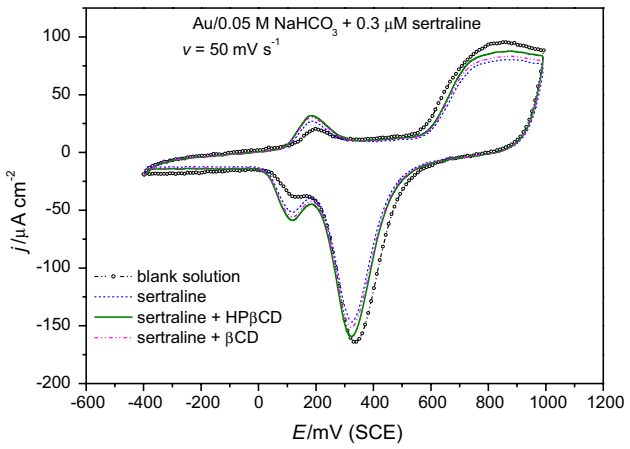

Fig. 2 CVs of Au electrode with 0.3 μM sertraline (dash line), inclusion complex of sertraline with HPβCD (full line), inclusion complex of sertraline with βCD (dash dot line) and in blank solution (dot line) in 0.05 M $NaHCO_3$. Scan rate 50 mV s^{-1}

It was published that the electrochemical oxidation of sertraline on a rutine/glassy carbon electrode (GCE) is an irreversible 2e/2H^+ reaction [9]. The deprotonation of the sertraline molecule proceeds at the cyclohexane unit at α carbon to methyl amino group. It seems that the formation of the inclusion complex of the sertraline with HPβCD leads to an easier deprotonation during electrochemical oxidation. At more positive potentials ($E > 450$ mV), anodic currents for the sertraline and its inclusion complex with HPβCD oxidation decrease in regard to the Au electrode in blank solution. It shows their inhibiting effect on the Au oxide formation obviously attributed to the presence of sertraline and diminished during complexation with HPβCD. The peak of oxide reduction also decreases in comparison to the oxide reduction of the Au electrode in a blank solution. The same electrochemical response was obtained when sertraline is complexed with βCD, while the reaction currents are somewhat lower in regard to inclusion complex with HPβCD.

For quantitative determination of sertraline, the SWV technique was employed. Figure 3a showed SWV curves of sertraline presented with one peak at ~100 mV. For sertraline concentrations higher than 0.5 μM, reaction currents decline due to the surface saturation with reaction species. The inclusion complex of sertraline with HPβCD depicts higher reaction currents in SWV measurements (Fig. 3b), as it was presented in the potentiodynamic measurements. As was observed by CV, the improvement of the oxidation ability regarding inclusion complexes was reported and also for SWV determination of amlodipine [34] and arylazo pyridone dyes [35]. Also, the peak potential was shifted towards more positive potentials for ~50 mV in the presence of the inclusion complex while the range of investigated concentrations was wider.

Considering the inclusion complex of sertraline with βCD, SWV measurements showed the same response as it was obtained for the inclusion complex of sertraline with HPβCD but with lower peak reaction currents. The SWV response showed that the oxidation peak currents for the sertraline standard were linearly dependent on its concentration in the range from 0.1 to 0.5 μM as it is presented in Fig. 4a. The obtained regression equation is j_p/μA $cm^{-2} = 31.33 + 4.70\ c$/μM ($R^2 = 0.999$, SD = 0.032 μA cm^{-2}) and LOD and LOQ are 2.0×10^{-8}, and 6.7×10^{-8} M, respectively. On the other hand, the oxidation peak of the inclusion complex of sertraline with HPβCD was linearly related to the concentration in the range 0.1–0.9 μM (Fig. 4b) giving the regression equation j_p/μA $cm^{-2} = 40.60 + 25.55\ c$/μM ($R^2 = 0.999$, SD = 0.22 μA cm^{-2}) and LOD and LOQ are 2.6×10^{-8} and 8.8×10^{-8} M, respectively. The regression equation obtained for the inclusion complex of the sertraline with βCD is j_p/μA $cm^{-2} = 25.925 + 22.75\ c$/μM ($R^2 = 0.999$, SD = 0.20 μA cm^{-2}) with a LOD and a LOQ being 2.6×10^{-8} and 8.6×10^{-8} M, respectively. The LOD and LOQ values

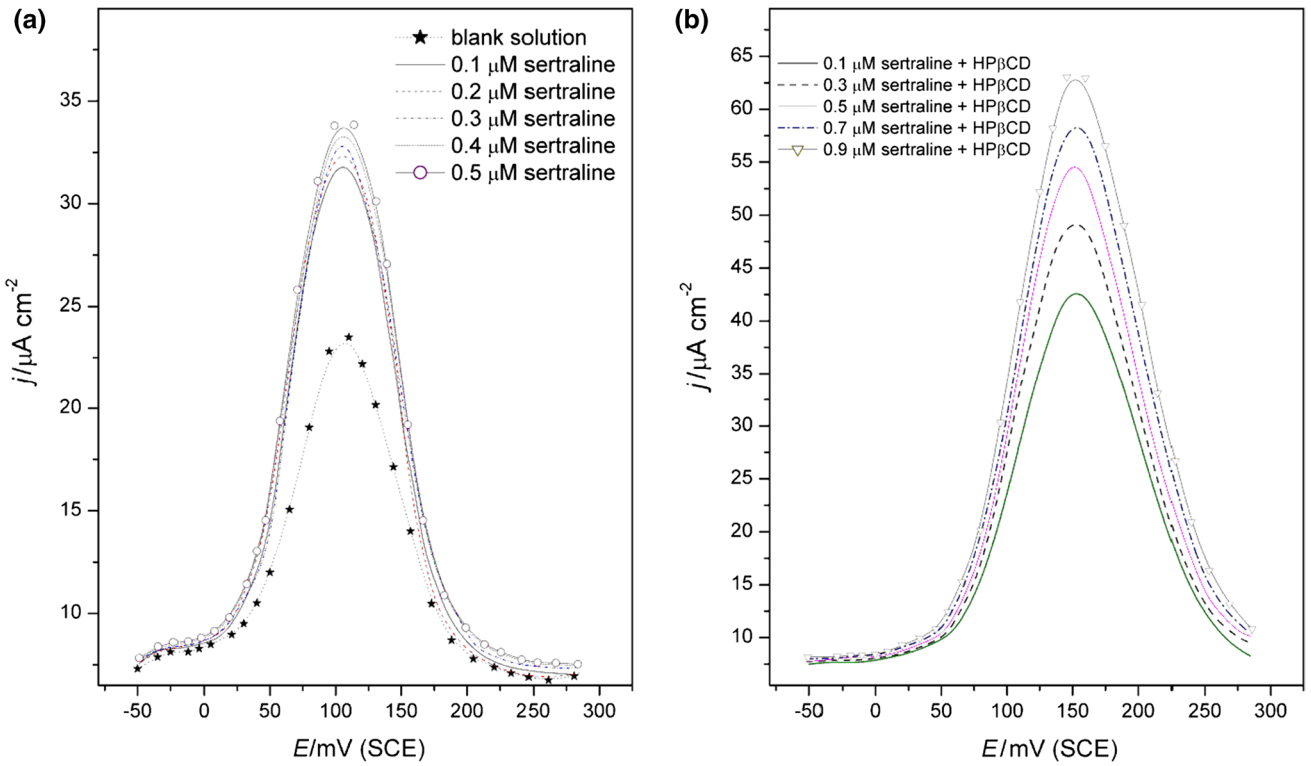

Fig. 3 Sertraline (**a**) and its inclusion complex with HPβCD (**b**) determined by SWV at Au (the concentrations added in electrolyte are presented in legend). Step size 5 mV, pulse size 25 mV and scan rate 10 mV s^{-1}, accumulation time 30 s, at the potential (− 50 mV)

were estimated from the regression lines as: LOD = 3 × SD/slope, and LOQ = 10 × SD/slope.

Detailed statistical parameters of obtained calibration curves are summarized in the Table 1. Linearity of a signal/concentration dependence was statistically confirmed using several approaches. The first approach was based on a visual inspection of residuals [36]. No presence of any trends, neither curvilinear, linear descending, linear ascending, nor the heteroscedasticity, i.e., increase in the variability of residuals with the increase of the analyte's concentration was observed which confirms the linearity of regression lines. The second approach was based on testing the conformity of the values of the slope calculated from each calibration point to the ±5% tolerance deviations from the average value of the slope [37]. For all three regression equations, the ±5% tolerance ranges of the slopes are: 4.47–4.94, 21.61–23.88, and 24.14–26.68 respectfully. In all three cases, all the points yield the slope values within these limits, which prove the linearity in the tested concentration range. The third way was based on comparison of adjusted R^2 values obtained in the case of linear regression and those obtained for the curvilinear models, such as quadratic and cubic polynomials [36]. The highest R^2_{adj} values, obtained in the case of simple linear models, prove that the linear models are the best fit option (Table 1). The last approach was based on a comparison of the variances of residuals of the linear fit versus the fit of higher orders, such as quadratic or cubic ones, by the means of the *F*-test (denoted as $F_{lin/quad}$, and $F_{lin/cub}$, respectively). In the case that the quadratic or cubic polynomials demonstrate a better fit into the calibration data, the *F* value should be statistically significant at a predefined significance level. Since no such statistical significance was observed in all three calibration models (Table 1), the linear fit was proven to be the best option. Comparing the regression equations, it can be noticed that the sensitivity in the presence of the inclusion complex was nearly 5 times higher.

Table 2 illustrates the obtained values of several important analytical parameters, such as linear range, LOD, LOQ, for the determination of sertraline that can be found in earlier reported papers. Comparing the obtained results on the Au electrode to previously published results presented in Table 2, concerning the electrochemical behaviour of sertraline, it can be concluded that an extension of the linear concentration range was accomplished [6–10]. The inclusion complexes of sertraline with HPβCD demonstrated a new electrochemical strategy for increasing the electrode's sensitivity in its quantitative determination. In addition, the obtained values of LOD and LOQ for the inclusion complex of the sertraline with HPβCD are comparable [10, 11, 38] or more than one order of magnitude

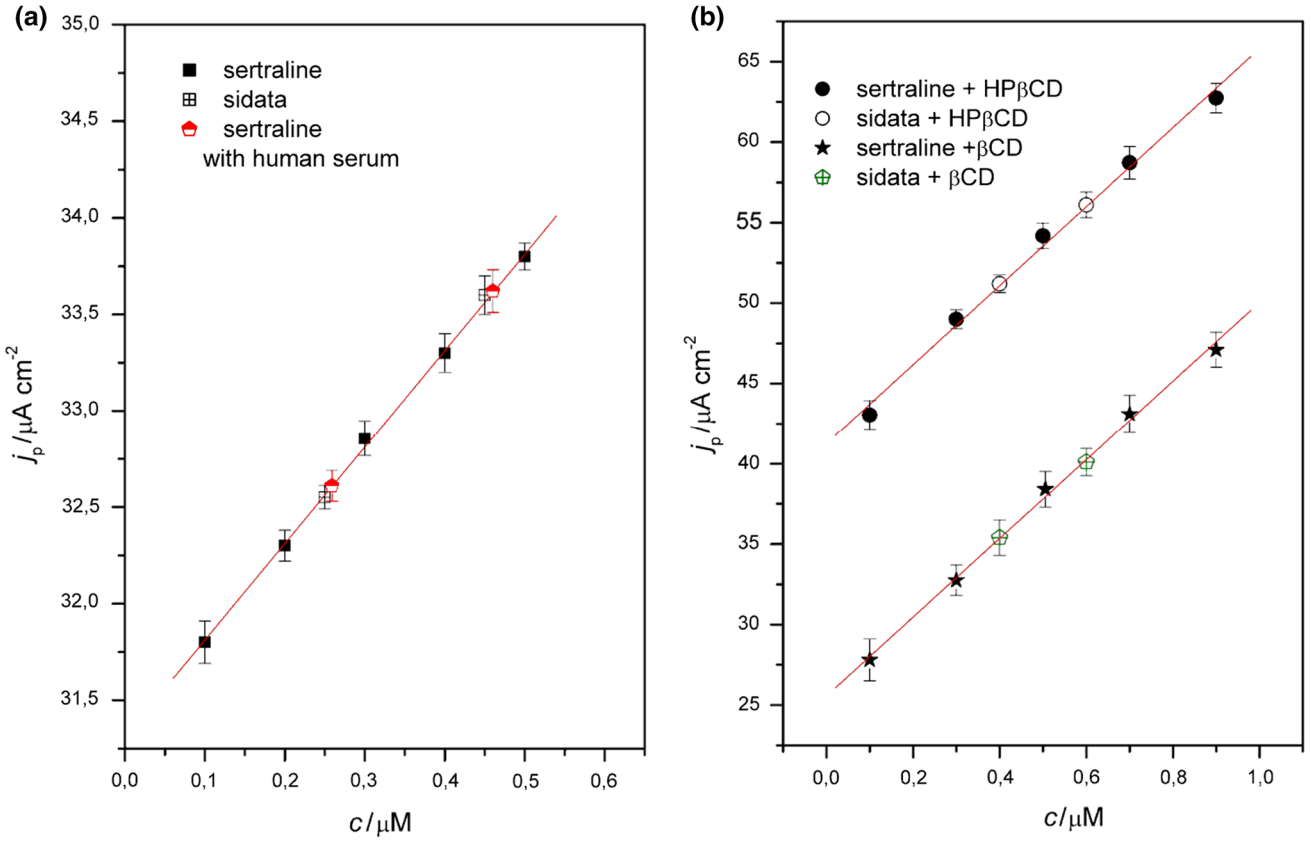

Fig. 4 Dependence of j_p from c (data collected from Fig. 3a) for the sertraline standard and its unknown concentrations in Sidata tablets and in human serum spiked with standard (**a**); and (data collected from Fig. 3b), for the inclusion complex of the sertraline standard with HPβCD and βCD

Table 1 Statistical parameters of obtained calibration models

Calibration model for SWV method	A	B (βCD)	B (HPβCD)
Intercept, a/μA cm^{-2}	31.33	25.92	40.62
Slope, b/μA cm^{-2} μM	4.70	22.75	25.55
Standard error of the intercept, s_a	0.033	0.18	0.20
Standard error of the slope, s_b	0.10	0.31	0.35
SD	0.032	0.20	0.22
R^2	0.9986	0.9994	0.9994
F	966.8	5400.7	5187.8
p	$2.1{\cdot}10^{-5}$	$5.6{\cdot}10^{-6}$	$5.9{\cdot}10^{-6}$
n	5	5	5
$R^2_{adj(lin)}$	0.9982	0.9993	0.9992
$R^2_{adj(q)}$	0.9979	0.9989	0.9980
$R^2_{adj(cub)}$	0.9977	0.9983	0.9980
$F_{lin/quad}$	0.88	0.67	0.38
$p(F)$	0.572	0.645	0.782
$F_{lin/cub}$	0.00	0.43	0.38
$p(F)$	0.9999	0.7756	0.7973

Table 2 Comparison of analytical parameters of various electrodes for the determination of sertraline

Electrode	Linear range/M	LOD/M	LOQ/M	Reference
HMDE	2.33×10^{-7}–3.15×10^{-6}	1.98×10^{-7}	–	[6]
HMDE	0.20×10^{-6}–1.20×10^{-6}	1.5×10^{-7}	5.0×10^{-7}	[7]
GC	4×10^{-5}–10^{-4}	1.04×10^{-5}	3.72×10^{-5}	[8]
R/GC	3×10^{-6}–9×10^{-5}	1×10^{-6}	–	[9]
Ni(II)-LD/Au	5×10^{-8}–5.5×10^{-6}	9.5×10^{-8}	–	[10]
CPE	1.99×10^{-7}–1.38×10^{-5}	2.23×10^{-8}	7.42×10^{-8}	[11]
Pencil lead	10^{-7}–10^{-6}	4.5×10^{-8}	–	[38]

higher than the published [6–8] as can be seen in Table 2. The evaluated parameters indicated a significant electrochemical response and possibility for analytical use. SWV measurements were further utilized for the quantitative determination of sertraline in pharmaceutical formulation as Sidata tablets.

Using the calibration curves (Fig. 4a, b), the values of unknown sertraline concentrations in Sidata tablets as well as in the human serum spiked with sertraline standard were determined. Prior to SWV investigations, each excipient as a content of pharmaceutical formulation Sidata was tested under the experimental conditions used in potentiodynamic measurements and all of them were electrochemically inactive.

The inclusion complexes of sertraline and Sidata with βCD were also investigated and the activity was compared to the results of sertraline and Sidata in the complex with HPβCD (Fig. 4b). The lower peak currents were obtained using βCD and the difference of the peak current of sertraline in the complex with HPβCD and βCD according to Fig. 4b was nearly 1.5 times. The observed electrochemical properties of sertraline in the complex with HPβCD or βCD were attributed to the influence of the structural parameters of cyclodextrins [39]. It has been shown that the presence of the hydroxypropyl groups additionally stabilizes the complex and appropriate interactions were in greater extent than in the case of βCD. The stronger binding ability concerning the inclusion complex of the azo dye with the HPβCD in regard to βCD has already been reported [35].

The accuracy of the proposed method was examined by recovery analysis applied for the set of sertraline concentrations as is presented in Table 3. The recovery, as an average value of 3 replicates varies from 96.74% to 102.60% and the relative standard deviation (R.SD) varies from 1.53% to 4.91%. This result indicates that the proposed electrochemical methods are reliable electroanalytical procedures for the quantification of sertraline. However, the lower relative standard deviations are obtained in the case of HPβCD and βCD added (1.64–2.59% and 1.53–1.68%, respectively) when compared to the method without the CD addition (4.73–4.91%).

For the quantitative detection of sertraline, the HPLC technique was also used. The linear response of the peak area to its concentration in the solution was shown in the range of 0.05–8 μM. The obtained regression equation is $y = 1824.887 + 0.84398\ x$ ($R^2 = 0.999$, SD = 1.06) and LOD and LOQ are 3.0×10^{-8} and 10.0×10^{-8} M, respectively.

All SWV determined sertraline concentrations were confirmed by HPLC–UV and the comparison with SWV is presented in Table 4. The comparison of sertraline concentration in the Sidata tablet and its inclusion complexes determination by SWV to HPLC method was performed by the *t*-test for two samples assuming equal variances. The obtained *t*-values for the SWV methods A, B (HPβCD) and B (βCD) (1.876, 0.155, and 0.914, respectively) were lower than the critical *t*-value (2.23 for the two-tailed test at the predefined $p = 0.05$ significance level), indicating that there is statistically no significant difference between the electrochemical and the HPLC method.

Conclusion

The obtained results showed that the gold electrode displayed a significant electrocatalytic response to the inclusion complexes of cyclodextrin with sertraline as the standard and Sidata tablets. Using and developing the SWV method and comparing it to the previously published results on different electrode surfaces concerning electrochemical behavior of sertraline, it can be concluded that the extension of

Table 4 The determination of sertraline in Sidata tablet using the proposed SWV methods

Method	Amount found (± SD)[a]/mg
SWV A	51.9 (± 2.5)
SWV B (HPβCD)	49.9 (± 1.0)
SWV B (βCD)	50.5 (± 1.1)
HPLC	50.0 (± 0.82)

[a]Arithmetic mean and standard deviation of six replicates

Table 3 Recovery studies of sertraline

SWV method	Amount added[a]/μM	Amount found/μM	Recovery/%	Relative standard deviation[b]/%
A	0.25 (tablet powder)	0.24	96.74	4.91
	0.45 (tablet powder)	0.46	102.60	4.73
	0.25 (serum)	0.26	102.41	4.91
	0.45 (serum)	0.44	97.87	4.73
B (HPβCD)	0.40 (tablet powder)	0.41	102.59	2.59
	0.60 (tablet powder)	0.60	99.27	1.64
B (βCD)	0.40 (tablet powder)	0.41	102.29	1.68
	0.60 (tablet powder)	0.61	102.14	1.53

[a]Solution of sertraline

[b]Number of replicates = 3

the linear concentration range was accomplished. Based on the constructed and statistically validated calibration curve, the values of unknown sertraline concentration in the Sidata tablet and in human serum spiked with the standard were determined opening perspectives for serious analytical applications.

Experimental

Sertraline hydrochloride, as the reference standard, and commercial drug Sidata were kindly provided by Hemofarm Stada A.D. (Vršac, Serbia). HPβCD (97.5%) and βCD (98%) were purchased from Sigma-Aldrich and Merck, respectively. All chemicals used were of p.a. grade. The deionized water is obtained by a Millipore Waters Milli-Q purification unit and used in all experiments.

Stock solution of the sertraline standard was prepared by dissolving the compound (2.918 μmol dm^{-3}) in deionized water. The stock solution of Sidata was prepared by powdering 10 tablets (50 mg of active compound) in a mortar. The adequate weight was measured and dissolved in deionized water under ultrasonic. After filtration through a filter paper, the solution was diluted with the deionized water to 100 cm^3. Both stock solutions were stored in a refrigerator at 4 °C. The inclusion complexes of sertraline and Sidata were prepared with HPβCD and βCD in molar ratio of 1:1. First, cyclodextrin solutions equimolar to drug stock solutions were prepared. Aliquots of equimolar solutions of the host and guest molecules were mixed in $NaHCO_3$ solutions to obtain a specified concentration of inclusion complexes. The obtained solutions were mixed for 24 h and allowed to equilibrate at room temperature over night before their addition to the electrochemical cell.

Human blood was collected from ten healthy volunteers and the serum was clinically prepared and spiked with sertraline standard in laboratories of Faculty of Medicine, University of Belgrade.

Electrochemical measurements were performed using PGZ 402 Volta Lab (Radiometer Analytical, Lyon, France) in a three-electrode cell with Au as the working electrode (surface area 0.5 cm^2), Au wire auxiliary electrode, and a calomel reference electrode (SCE). The Au working electrode was polished with diamond paste, cleaned with a mixture of 18 MΩ water and sulfuric acid and further cleaned with 18 MΩ deionized water in an ultrasonic bath. The electrolytes were deoxygenated by purging with nitrogen. The working electrode was checked prior to each experiment by cycling the potential scan between -400 and 1000 mV in a supporting solution (0.05 M $NaHCO_3$, pH 8.4) at the scan rate of 50 mV s^{-1} until the unchanged CV characteristics for the Au electrode were achieved. The accumulation of the samples at the Au electrode was carried out for 30 s at -50 mV. After that, SWV initiated in the positive potential direction was performed. The following parameters were used as the optimum to record the SWVs: step size 5 mV, pulse size 25 mV, frequency 2 Hz, scan rate 10 mV s^{-1}.

The HPLC measurement was performed on Agilent 1100 HPLC system consisting of Diode Array Detector, binary pump, thermostated column compartment, autosampler, and a micro vacuum degasser. HPLC chromatographic separation was carried out on Zorbax Eclipse XDB C-18 column (150×4.6 mm, 5 μm) using isocratic elution with the mobile phase consisting of a buffer solution (2.88 g of sodium dodecyl sulfate, 3.40 g of tetra-*n*-butyl ammonium hydrogen sulfate, and 2.76 g of sodium phosphate monobasic monohydrate and transfer in 1000 cm^3 water), methanol, and acetonitrile in a 40:40:30 ratio. Flow rate was 2 cm^3 min^{-1}, the injection volume was set at 20 mm^3, the column temperature was 40 °C and UV detection was carried out at 220 nm.

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